

✓
Lonberg and Kay
Application No.: 09/724,965
Page 2

PATENT

7 segment, at least one J_k segment, at least one human C_k segment, and a human 3' kappa
8 enhancer segment.

1 18. The construct of claim 17, wherein the human 3' kappa enhancer
2 segment is a 4 kb BamHI fragment containing the human 3' kappa enhancer.

1 19. A transgenic nonhuman mammal comprising the transgene of
2 claim 1.

1 20. The transgenic nonhuman mammal of claim 19, wherein the
2 transgene is expressed in B cells of the transgenic nonhuman mammal.

1 21. The transgenic nonhuman mammal of claim 19, wherein the
2 transgene is in the germline of the transgenic non-human mammal.

1 22. The transgenic nonhuman mammal of claim 19, further comprising
2 an Ig heavy chain transgene construct.

1 23. The transgenic nonhuman mammal of claim 21, wherein the
2 transgene is rearranged.

1 24. The transgenic nonhuman mammal of claim 21, wherein the
2 transgene is unrearranged.

1 25. The transgenic nonhuman mammal of claim 22, wherein the
2 transgene is rearranged.

1 26. The transgenic nonhuman mammal of claim 22, wherein the
2 transgene is unrearranged.

1 27. The transgenic nonhuman mammal of claim 19, wherein the
2 mammal makes an antibody response following immunization with an antigen.

1 28. The transgenic nonhuman mammal of claim 27, wherein the
2 antigen is a human antigen.

1 29. The transgenic nonhuman mammal of claim 27, wherein the
2 antibody response comprises a population of antibodies which comprise human μ chain-
3 containing immunoglobulins and human γ chain-containing immunoglobulins.

1 30. The transgenic nonhuman mammal of claim 20, wherein the B
2 cells produce a heterologous antibody.

Lonberg and Kay
Application No.: 09/724,965
Page 3

PATENT

1 31. The transgenic nonhuman mammal of claim 30, wherein the B
2 cells produce a population of heterologous antibodies of more than one isotype.

1 32. The transgenic nonhuman mammal of claim 19 wherein the
2 nonhuman mammal is a rodent.

1 33. A method for generating a plurality of B cells expressing human
2 antibody sequences, the method comprising:
3 providing a transgenic nonhuman mammal of claim 19; and
4 immunizing the transgenic nonhuman mammal to generate B cells
5 producing a population of heterologous antibodies.

1 34. The method of claim 33, further comprising collecting the B cells
2 producing a population of heterologous antibodies.

1 35. The method of claim 34, further comprising fusing the B cells
2 producing a population of heterologous antibodies with immortalized cells to form
3 hybridomas.

1 36. The method of claim 35 further comprising collecting the human
2 antibody sequences from the hybridomas.

1 37. The method of claim 36, wherein the human antibody sequences
2 are purified.

1 38. The method of claim 33, further comprising collecting the
2 sequences encoding human antibodies.

1 39. The method of claim 38, wherein the sequences encoding human
2 antibodies are full length.

1 40. The method of claim 39, further comprising expressing the
2 sequences in a transfected cell.

1 41. A method of generating antigen-specific hybridomas secreting
2 human sequence antibody, the method comprising:
3 immunizing the transgenic nonhuman mammal of claim 19 with a
4 predetermined antigen;

Lonberg and Kay
Application No.: 09/724,965
Page 4

PATENT

5 fusing lymphocytes from the transgenic mouse with immortalized cells to
6 form hybridoma cells; and

7 determining the binding of the antibody produced by the hybridoma cells
8 to the predetermined antigen.

1 42. A method for generating a human sequence antibody that binds to
2 a predetermined antigen, the method comprising the following steps:

3 immunizing the transgenic nonhuman mammal of claim 19 with a
4 predetermined antigen; and

5 screening hybridoma cells formed for the presence of antigen reactive
6 antibodies.

1 43. The method of claim 42, wherein the antigen reactive antibodies
2 are secreted from the hybridoma in culture.

1 44. The method of claim 42, wherein the antigen reactive antibodies
2 are substantially pure.

1 45. A method for producing rearranged immunoglobulin sequences
2 comprising:

3 providing the transgenic nonhuman mammal of claim 19;

4 obtaining the rearranged immunoglobulin sequences from the transgenic
5 nonhuman mammal.

1 46. The method of claim 45, wherein the obtaining step comprises
2 collecting B cell lymphocytes containing the rearranged immunoglobulin sequences from
3 the transgenic nonhuman mammal.

1 47. The method of claim 46, wherein the obtaining step comprises
2 isolating and amplifying mRNA from B cell lymphocytes to generate cDNA.

1 48. The method of claim 47, further comprising isolating and
2 amplifying heavy and light chain variable region sequences from the cDNA.

1 49. An isolated nucleic acid encoding the heavy and light chain
2 variable region sequences of claim 48.

Lonberg and Kay
Application No.: 09/724,965
Page 5

PATENT

1 50. An isolated nucleic acid encoding the heavy chain variable region
2 sequences of claim 48.

1 51. An isolated nucleic acid encoding the light chain variable region
2 sequences of claim 48.

1 52. A vector comprising the nucleic acid of claim 49.

1 53. An expression vector comprising the nucleic acid of claim 49 in
2 which the heavy and light chain variable regions sequences of the nucleic acid are
3 operatively linked with a regulatory sequence that controls expression of the nucleic acid
4 in a host cell.

1 54. A host cell comprising the nucleic acid of claim 49, or progeny of
2 the cell.

1 55. The host cell of claim 54 which is a eukaryote.

1 56. The method of claim 45, further comprising:
2 culturing the host cell of claim 54 under conditions such that the nucleic
3 acid is expressed; and
4 recovering the nucleic acid from the cultured host cell or its cultured
5 medium.